Optimal mode of operation for biomass production

Betlem, Ben H.L.; Roffel, Brian; Mulder, P.

Published in:
Chemical Engineering Science

DOI:
10.1016/S0009-2509(02)00149-5

PERGAMON

Chemical Engineering Science 57 (2002) 2799–2809

Chemical Engineering Science

www.elsevier.com/locate/ces

Optimal mode of operation for biomass production

Ben H. L. Betlem[a,*], Pieter Mulder[b,1], Brian Roffel[a]

[a] *Department of Chemical Engineering, University of Twente, P.O. Box 217, 7500 AE Enschede, The Netherlands*
[b] *Department of Applied Physics, University of Twente, P.O. Box 217, 7500 AE Enschede, The Netherlands*

Received 29 March 2001; received in revised form 15 November 2001; accepted 29 March 2002

Abstract

The rate of biomass production is optimised for a predefined feed exhaustion using the residue ratio as a degree of freedom. Three modes of operation are considered: continuous, repeated batch, and repeated fed-batch operation. By means of the Production Curve, the transition points of the optimal modes of operation are derived. The analytical expressions of these transitions for variable bioreaction kinetic parameters are determined. The key measures "degree of difficulty of conversion" and "degree of exhaustion" are introduced to define the optimal modes in more general terms. The "degree of difficulty" describes the effect of the kinetic parameters and the feed substrate concentration on the conversion; the "degree of exhaustion" describes the desired final condition. In fed-batch operation, the proposed constant feed policy approximates the optimal feed policy closely.

Keywords: Bioreactor; Optimal operation; Batch operation; Fed-batch operation; Cyclic operation; Repeated operation

1. Introduction

For biomass production three basic modes of operation are possible: continuous, batch or fed-batch operation. Continuous operation is usually not the most productive option as sufficiently exhaustion of the enriched feed can only be realised at rather low feed rates. In the batch and fed-batch mode, a part of the bioculture mass can be left in the reactor for the next batch, causing a cyclic operation of repeated batches or fed-batches. The residue, defined as a fraction of the maximum reactor volume, is a degree of freedom for optimisation.

Many studies have been published about the optimal control of biomass and metabolic production. Some articles are dedicated to a single fed-batch run and pay special attention to the use of Pontryagin's maximum principle or Green's theorem (San & Stephanopoulos, 1984, 1986; Cazzador, 1988; Park & Ramirez, 1988). The solution is said to be singular in cases where the maximum principle does not lead to a well-defined relation between the state and the control variable. Menawat, Muthurasan and Coughanowr (1987) and Palanki, Kravaris, and Wang (1993) derived exact solutions for the singular solution, Modak, Lim, and Tayeb (1986) determined the optimal feeding policies for different metabolic kinetic models. Parulekar (1992) examined the admissibility and sensitivity of a singular control for a vast range of fermentation types. Early studies concerning cyclic batch or cyclic fed-batch biomass production are by Pirt (1974), Keller and Dunn (1978), Weigand (1981) and Weigand, Lim, Creagan, and Mohler (1979). The last mentioned authors optimised fed-batch cyclic operation for a prescribed final biomass and feed concentration. The time optimal feeding policies were based on fixed values for the residue volume. However, this value is a variable that can be chosen to maximise the productivity. The maximal production is most readily found by a trial-and-error procedure. Based on experimental work many groups report productivity improvements by using cyclic fed-batch, e.g. Lee, Pham, Weigand, Harvey and Bentley (1996), Chang, Ryu, Park, and Kim (1998). Matsubaru, Hasegawa, and Shimizu (1985) and Hasegawa, Matsubara, and Shimizu (1987) studied the optimal cyclic operation as a two-objective programming problem for biomass production and a three-objective problem for metabolite production. In a two-objective

* Corresponding author.
E-mail address: b.h.l.betlem@ct.utwente.nl (B. H. L. Betlem).
[1] Presently with: Department of Chemical and Process Engineering, University of Newcastle, Merz Court, Newcastle upon Tyne, NE1 7RU, UK

PII: S0009-2509(02)00149-5

programming problem the productivity together with the conversion are optimised. Shukla and Pushpavanam (1998) determined an optimal numerical strategy using constraints. Cooper and co-workers (Brown & Cooper, 1991; Wincure, Cooper, & Rey, 1995; Hughes & Cooper, 1996; Godin, Cooper, & Rey, 1999) use a cyclic operation, called self-cycling fermentation, with a fixed residue ratio of 50% to study the kinetic parameters of several fermentation processes under dynamic circumstances. The periodicity is determined by a growth-associated parameter. The fixed ratio will rarely result in an optimal production rate. Modak and Lim (1992) systematically analysed the mode of operation for optimal metabolite productivity with the inlet and outlet flow as control variables. The optimal mode depends on the ratio of the metabolite production rate and the substrate consumption rate. For a ratio which either increases, decreases, goes through maximum or remains constant with increasing substrate ratio, the optimal modes are single-cycle batch, continuous, single-cycle fed-batch and repeated batch, respectively.

With exception of the last, all studies discuss the determination and often the realisation of an optimal control trajectory for a specific conversion and mode of operation. The mode of operation of a bioreactor is fixed a priori and the optimal policies determined accordingly. In this article the mode itself is subject of discussion. An analytical solution of the transition points according to Pontryagin's Maximum Principle is not simple as the optimisation criteria and the process behaviour for continuous and cyclic operation are not equal and only a numerical solution with regard to the residue volume is possible. In this article the optimal mode are determined from analytical descriptions of the transition points derived from the Production Curve. By varying the kinetic parameters, the feed composition and the final exhaustion requirement over a wide range, more general rules can be obtained.

This study of bioreactions restricts itself to substrate inhibited production of biomass. Monod kinetics is considered to be a limit case of substrate-inhibition kinetics. First, the behaviour of biomass production as a function of the kinetic parameters and the goals of the operation are described. Based on this behaviour, the different control policies for a single cycle are introduced. Then, the following new items will be discussed:

- The optimal control mode is determined for the total range of the kinetic parameters, feed concentrations, and required final concentrations.
- The transition points between the optimal modes of operation are analytically defined.
- Key measures for the required exhaustion and the process capability for exhaustion are introduced to describe the areas of optimal operation in a more general way.
- A sub-optimal fed-batch control policy is introduced, since this operation is easier to implement, and is compared with the optimal feed policy.

2. Process behaviour and operation goals

Substrate is converted by the biomass into additional biomass. It is assumed that the reactor is ideally mixed. The biomass and substrate are represented by their concentrations in the culture, called X and S, respectively. The generalised component balances for the well-mixed bioreactor are

$$\frac{\mathrm{d}X}{\mathrm{d}t} = \mu\{S\}X - \frac{F_{\mathrm{in}}}{V}X, \tag{1}$$

$$\frac{\mathrm{d}S}{\mathrm{d}t} = -\frac{\mu\{S\}}{Y}X + (S_F - S)\frac{F_{\mathrm{in}}}{V}, \tag{2}$$

$$\frac{\mathrm{d}V}{\mathrm{d}t} = F_{\mathrm{in}} - F_{\mathrm{out}}, \tag{3}$$

where Y is the biomass yield and $\mu\{S\}$ is the specific growth rate, which depends on S. Y can also be a function of S, but in this work, Y is assumed constant. In addition, it is assumed that both biomass decay and maintenance requirements are negligible. The growth rate $\mu\{S\}$ relates the change in biomass concentration to the substrate concentration. Roels (1983) lists many kinetic models. Two types of relationships for $\mu\{S\}$ are commonly used: the substrate saturation model (Monod equation) and the substrate inhibition model). Models with more than three parameters are difficult to determine and do often not perform better (Edwards, 1970). From cyclic batch experimental work it appears that a rather simple kinetic model suffices (Lee et al., 1996; Hughes & Cooper, 1996). Substrate inhibited growth can be described by

$$\mu\{S\} = \mu_{\max}\frac{S}{K_S + S + S^2/K_I} \tag{4}$$

where K_S is the saturation or Monod constant, K_I is the inhibition constant and $\mu_{\max}$ is the maximum specific growth rate. The value of K_S expresses the affinity of the biomass for the substrate. The Monod growth kinetics is a special case of the substrate inhibition kinetics. Its equation is derived from Eq. (4) when the inhibition term in the denominator is neglected, thus when $K_{I\to\infty}$. In Fig. 1, the dependence of the substrate concentration on the saturated and the substrate inhibited growth is shown. For strongly inhibited growth, at higher substrate concentrations the growth rate decreases significantly. Substrate inhibited growth rate has a maximum for $S = S_{\mathrm{opt}}$:

$$S_{\mathrm{opt}} = \sqrt{K_I K_S}. \tag{5}$$

The goal of this work is to maximise the biomass production rate. For continuous operation the specific production rate, which is the production rate per unit of volume, is defined by

$$PR_{\mathrm{continuous}} = X_f F_{\mathrm{in}}/V_{\max}, \tag{6}$$

where X_f is the biomass product concentration, F_{in} is the feed rate and $V_{\max}$ is maximum reactor volume.

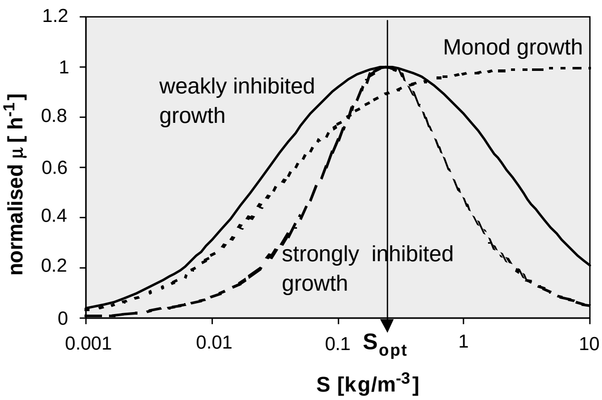

Fig. 1. Growth curves: Monod (K_S = 0.03 (kg m^{-3})), weak inhibition (K_S = 0.03 (kg m^{-3}), K_I = 2 (kg m^{-3})), strong inhibition ($K_S = 3$ (kg m^{-3}), $K_I = 0.02$ (kg m^{-3})).

Using the stationary form of Eqs. (1) and (2) with $V = V_{\max}$ and $S = S_f$, this relation can be rewritten as a function of the variable S_f

$$PR_{\text{continuous}} = \mu\{S_f\}Y(S_F - S_f). \tag{7}$$

The relative production (rate) is defined as the specific production (rate) divided by the potential specific production:

$$P_{\text{potential}} = Y(S_F - S_f). \tag{8}$$

With this expression, it is possible to normalise the production for different values of substrate feed. Thus the relative specific production rate for a continuous operation becomes:

$$PR_{\text{continuous,relative}} = \mu\{S_f\}. \tag{9}$$

For cyclic operation, the specific production rate is:

$$PR_{\text{batch}} = \frac{X_f(V_f - V_0)}{T}\frac{1}{V_{\max}}, \tag{10}$$

where V_f and V_0 are the reactor volume before and after product removal, and T is the cycle time.

The constraints for the production optimisation are the substrate feed composition and the remaining substrate composition in the product, S_f.

3. Control policies

Three modes of operation have been studied. The application areas are shown in Table 1.

Fig. 2 illustrates the operation modes for inhibited growth. Fig. 2A and 2B also apply for Monod growth.

• Continuous operation (Fig. 2A): During continuous operation, all the three state variables, X, S, and V, are constant. For a maximum production rate, the volume remains at its maximum, $V_{\max}$. This operation is useful at relatively low exhaustion. When the substrate concentration, S_f, is fixed at its requirement, the feed rate is fixed and no degrees of freedom for optimisation remain.

Table 1
Application areas of modes of operation

Operation mode	Growth model	
	Monod	Substrate-inhibition
Continuous	Limited exhaustion	Limited exhaustion
Repeated batch	Extreme exhaustion	Extreme exhaustion weak inhibition
Repeated fed-batch	Not applicable	Extreme exhaustion strong inhibition

• Repeated batch operation (Fig. 2B): During the reaction, the volume is constant ($V_{\max}$) and the substrate concentration decreases. When the desired substrate concentration S_f has been reached, a part of the reactor contents is removed and enriched with fresh feed until S_b. The removed part is the product. The volume ratio, which is refreshed every cycle by new feed, is the only degree of freedom. For the filling and emptying rate of the cyclic operation, no maximum rate has been applied. It is assumed that the time involved is negligible. Actually, batch operation is a special case of fed-batch, applicable when a large exhaustion is combined with a relatively weak inhibition, which allows relatively high substrate concentrations.

• Repeated fed-batch operation: This kind of operation is only of interest in case of strongly substrate-inhibited growth, since the operation should be maintained around the maximum growth rate. After removal of a part of the reactor contents, this part is only partially refreshed by new feed, whereas the rest is fed subsequently according a certain control policy.

Two different types of fed-batch operation can be applied:

- Dynamic optimal feed rate (Fig. 2C): After product removal, refreshing with fresh feed is applied such that: $S_b = S_{\text{opt}}$. Then, the feed rate follows the singular arc: it keeps the substrate concentration at the point of maximum growth rate S_{opt} (Cazzador, 1988). The degrees of freedom for optimisation are the residue ratio and the time dependent feed rate.

- Sub-optimal constant feed rate (Fig. 2D): This is an approximation of the optimal feed policy. During the feeding phase the feed rate is kept constant. As a result, the substrate concentration will shift around the point of maximum growth rate. The degrees of freedom for optimisation are the residue ratio and the constant feed rate.

4. Optimal cyclic operation

Shukla and Pushpavanam (1998) proved that for a pseudo-stationary cyclic (fed-) batch operation the relationship between X and S develops into a static relationship. By dividing Eq. (1) by Y, multiplying Eq. (3) by ($-S_F$) and adding these two to Eq. (2) the following relationship

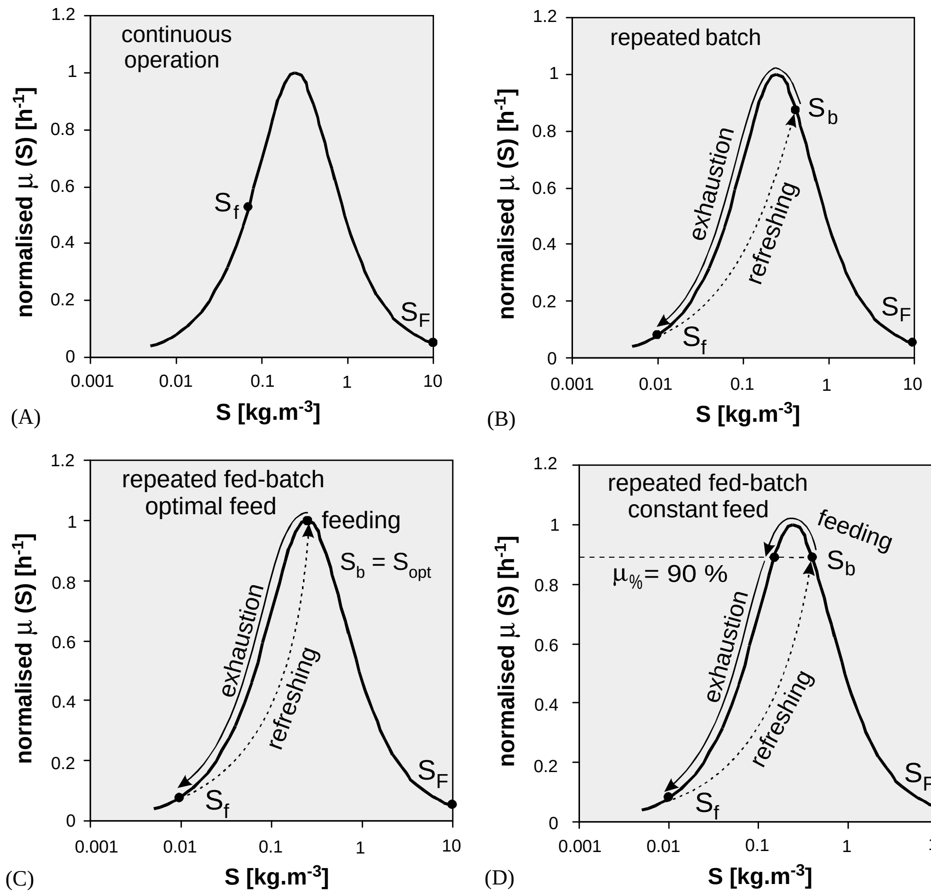

Fig. 2. Basic policies for inhibited growth: (A) continuous, (B) repeated batch, (C) repeated fed-batch operation with dynamic optimal feed rate, and (D) repeated fed-batch operation with constant feed rate.

is obtained:

$$\frac{\mathrm{d}}{\mathrm{d}t}\left(\frac{XV}{Y} - V(S_F - S)\right) = 0. \tag{11}$$

The repeated use of this relationship leads to

$$X = Y(S_F - S). \tag{12}$$

This is illustrated in Fig. 3 where the trajectories of the refreshing process and biomass conversion develop equal slopes. When η is small, the cycles will converge faster. Eq. (12) is an important observation and enables us to eliminate S in favour of X and reduces the state equations by one.

Like for continuous operation, using Eq. (12) with $V_f = V_{\max}$, the production rate Eq. (10) can be rewritten as a function of the state variable S_f:

$$PR_{\mathrm{batch}} = Y(S_F - S_f)(1-\eta)/T \quad \text{with } \eta = V_0/V_{\max}. \tag{13}$$

By using Eq. (8) the relative specific production rate for cyclic batch operation becomes

$$PR_{\mathrm{batch,relative}} = (1-\eta)/T. \tag{14}$$

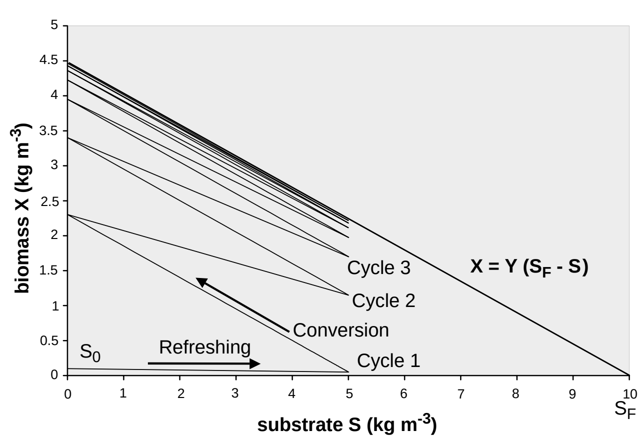

Fig. 3. Phaseplot of substrate and biomass for cyclic operation with refreshing and conversion steps.

The cycle time T depends on the control strategy and is derived by Matsubaru et al. (1985)

$$T_{RB} = \frac{1}{\mu_{\max}}\left[-\frac{(1-\eta)(S_F - S_f)}{K_I} + \frac{\mu_m}{\mu\{S_F\}}\ln\left(\frac{1}{\eta}\right) + \frac{K_S}{S_F}\ln\left(\frac{\eta S_f + (1-\eta)S_F}{S_f}\right)\right], \tag{15}$$

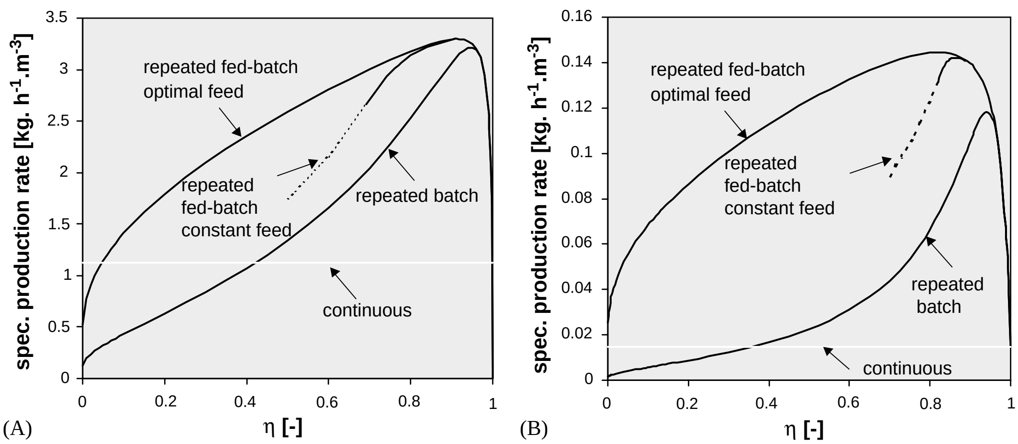

Fig. 4. Production for continuous, repeated batch, optimal and suboptimal (constant feed rate) repeated fed-batch mode ($\mu_{\max} = 1$ (h^{-1}), $Y = 0.45$, $S_F = 10$ (kg m^{-3}), $S_f = 0.01$ (kg m^{-3})). (A) weak inhibition ($K_S = 0.03$ (kg m^{-3}), $K_I = 2$ (kg m^{-3})), and (B) strong inhibition ($K_S = 3$ (kg m^{-3}), $K_I = 0.02$ (kg m^{-3})).

Table 2
Parameters of experimental data

Source	μ_m (h^{-1})	Y (kg/kg)	K_S (kg m^{-3})	K_I (kg m^{-3})	S_F (kg m^{-3})	S_f (kg m^{-3})
Wincure et al.	0.6	0.73	0.007	∞	0.65	0.03
Hughes, Cooper	0.774	0.625	0.001	0.670	1.94	Small

Table 3
Comparison of cycle time with experimental data

Source	Cycle time reported (min)	Calculated cycle time (min)	Reported conversion (kg h^{-1} m^{-3})	Expected maximal conversion (kg h^{-1} m^{-3})
Wincure et al.	72.3 ± 2	72.6	0.197	0.23
Hughes, Cooper	96 ± 4.5	97.7	0.60	1.25–1.50

$$T_{RFB} = \frac{1}{\mu_{\max}}\left[-\frac{(S_{\text{opt}} - S_f)}{K_I} + \frac{\mu_m}{\mu\{S_F\}}\ln\left(\frac{S_F - S_f}{S_F - S_{\text{opt}}}\right) + \frac{K_S}{S_F}\ln\left(\frac{S_{\text{opt}}}{S_f}\right) + \frac{\mu_m}{\mu\{S_{\text{opt}}\}}\ln\left(\frac{1}{\eta}\,\frac{S_F - S_{\text{opt}}}{S_F - S_f}\right)\right]. \tag{16}$$

It can be proven that continuous operation is a limit case of batch operation. Eq. (7) can be derived from Eq. (13) by taking the limit $\eta \to 1$, in which case $T \to 0$. The constraints for the production optimisation are the substrate feed concentration, S_F, and the remaining substrate concentration in the product, S_f.

Fig. 4 shows the production rates of four modes of operation for two specific sets of kinetic parameters. Continuous operation can be calculated analytically from Eq. (7), repeated batch from Eq. (13) combined with Eq. (15) and fed-batch from Eq. (13) with Eq. (16). The sub-optimal repeated fed-batch with constant feed rate has been simulated. Fig. 4A concerns weak inhibition and is similar to the example used by Weigand (1981) and Matsubaru et al. (1985). Fig. 4B reflects strong inhibition. The relative differences between the production rates of the different policies in the case of weak inhibition are smaller than in the case of strong inhibition.

The calculations of the cycle times and production rates are compared with the experimental data reported by Wincure et al. (1995) for the degradation of ethanol by *Acinetobacter calcoaceticus* RAG-1 and by Hughes and Cooper (1996) for the degradation of phenol by *Pseudomonas putida*. (see Tables 2 and 3). For both experiments cyclic batch operation has been used with a removal ratio of 0.5. By using higher ratios of 0.8 and 0.95, respectively, large improvements of the conversion can be expected.

Fig. 5 shows the maximum production rate and the residue volume ration η for a large range of the kinetic parameter

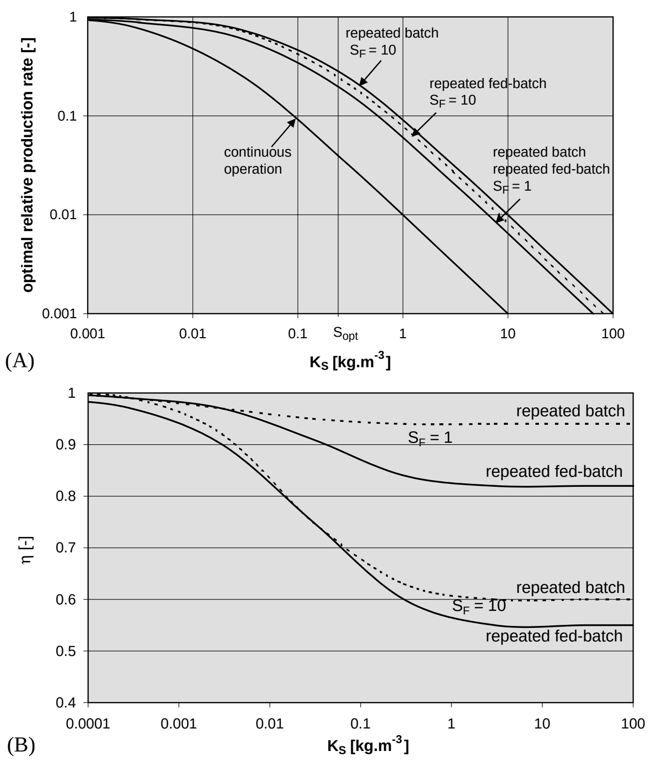

Fig. 5. Optimal relative production rate (A) and degree of freedom η (B) as a function of the kinetic parameter K_S (S_{opt} is constant, $\mu_{\max} = 1$ (h^{-1}), $Y = 0.45$, $S_F = 1,\ 10$ (kg m^{-3}), $S_f = 0.01$ (kg m^{-3})).

K_S for three operation modes. For $K_S \ll S_{\text{opt}}$, the η as well as the optimal relative production rate approaches one. In that region the optimal operation approximates continuous operation. For $K_S \gg S_{\text{opt}}$, η goes to a value independent of K_S and the production rate becomes inversely related to K_S as the cycle time depends linearly to K_S. This can be shown by substituting Eq. (4) for S_{opt} and (5) in Eqs. (15) and (16), which results in

$$T_{RB} = \frac{K_S}{\mu_m}\left[-\frac{(1-\eta)(S_F - S_f)}{S_{\text{opt}}^2} + \frac{S_F}{S_{\text{opt}}^2}\ln\left(\frac{1}{\eta}\right) + \frac{1}{S_F}\ln\left(\frac{\eta S_f + (1-\eta)S_F}{S_f}\right)\right], \tag{17}$$

$$T_{RFB} = \frac{K_S}{\mu_m}\left[-\frac{(S_{\text{opt}} - S_f)}{S_{\text{opt}}^2} + \frac{S_F}{S_{\text{opt}}^2}\ln\left(\frac{S_F - S_f}{S_F - S_{\text{opt}}}\right) + \frac{1}{S_F}\ln\left(\frac{S_{\text{opt}}}{S_f}\right) + \frac{2}{S_{\text{opt}}}\ln\left(\frac{1}{\eta}\,\frac{S_F - S_{\text{opt}}}{S_F - S_f}\right)\right]. \tag{18}$$

In case of high feed composition, fed-batch is preferable, as the process proceeds with substrate concentrations close to optimal growth composition S_{opt}. For $S_F = 10$ kg m^{-3} the difference in the relative production rate is about 20%. The residue volume ratio η at higher feed concentrations is, consequently, larger. For feed compositions close to and lower than S_{opt}, no difference occurs between repeated batch and repeated fed-batch operation.

5. Areas of optimal mode of operation

In this section, the optimal mode of operation is determined by means of the Production Curve. The curve, shown in Fig. 6, represents the specific production as a function of the cycle time for a specific control policy. The curve is suitable to determine the maximal performance of batch processes (Rippin, 1983). It can be calculated by simulations, but can also be derived from experiments when the parameters are unknown. To determine the optimal control mode for a particular final state, the required final state has been taken here as the starting point and therefore placed, for convenience, at the bottom left corner.

For the biomass production, it is assumed that $S_b > S_{\text{opt}}$ and $S_f < S_{\text{opt}}$. A batch cycle starts at $V(t_b) = V_{\max}$ and $S(t_b) > S_{\text{opt}}$ (at the upper right corner Fig. 6A). During exhaustion, first, the conversion rate increases and after passing the inflection point at S_{IP}, it decreases and is exhausted until $S(t_f) = S_f$ reached (at the lower left corner). According to Eq. (1), the biomass growth is maximal, when the product of $\mu\{S\}$ and X is maximal. Consequently, the maximum production rate does not coincide with the maximum growth rate: $S_{IP} < S_{\text{opt}}$. Since continuous operation is the limit case of batch operation for $T \to 0$, the slope of the tangent at the Production Curve equals the continuous production rate. A fed-batch cycle starts at $V(t_b) < V_{\max}$ and $S(t_b) = S_{\text{opt}}$. Following the singular arc, the growth rate remains the same until $V = V_{\max}$. From this point onward, the course of the fed-batch operation is the same as that of the batch operation.

The Production Curve has a S-shape with S_{opt} as the central point. From S_b to S_{opt}, the biomass grows exponentially, whereas from S_{opt} to S_f the substrate exhaustion determines the production. Fig. 6B shows the relative production. For $S \geqslant S_{\text{opt}}$, it appears that the fed-batch production is independent of the feed composition S_F, because the biomass production is linear to $(S_F - S_{\text{opt}})$. However, for batch operation with higher feed concentrations, smaller residue volume ratio are desirable and consequently the relative production decreases. Fig. 6C shows the absolute production for a particular case. The values of S along the Production Curve are indicated. For $S < S_{\text{opt}}$, the exhaustion proceeds equally for batch and fed-batch operation and is independent of the feed composition S_F or the final requirement S_f.

The optimal operation for a certain S_f can be found by taking the maximum tangent to the Production Curve in the point S_f. When the tangent to the Production Curve has a maximum in S_f itself, then continuous operation in S_f is optimal with a specific production rate of $\mathrm{d}P/\mathrm{d}t$. When the tangent contacts the curve in another point, then cyclic operation is optimal. In Fig. 6A the points S_b and S_f indicate

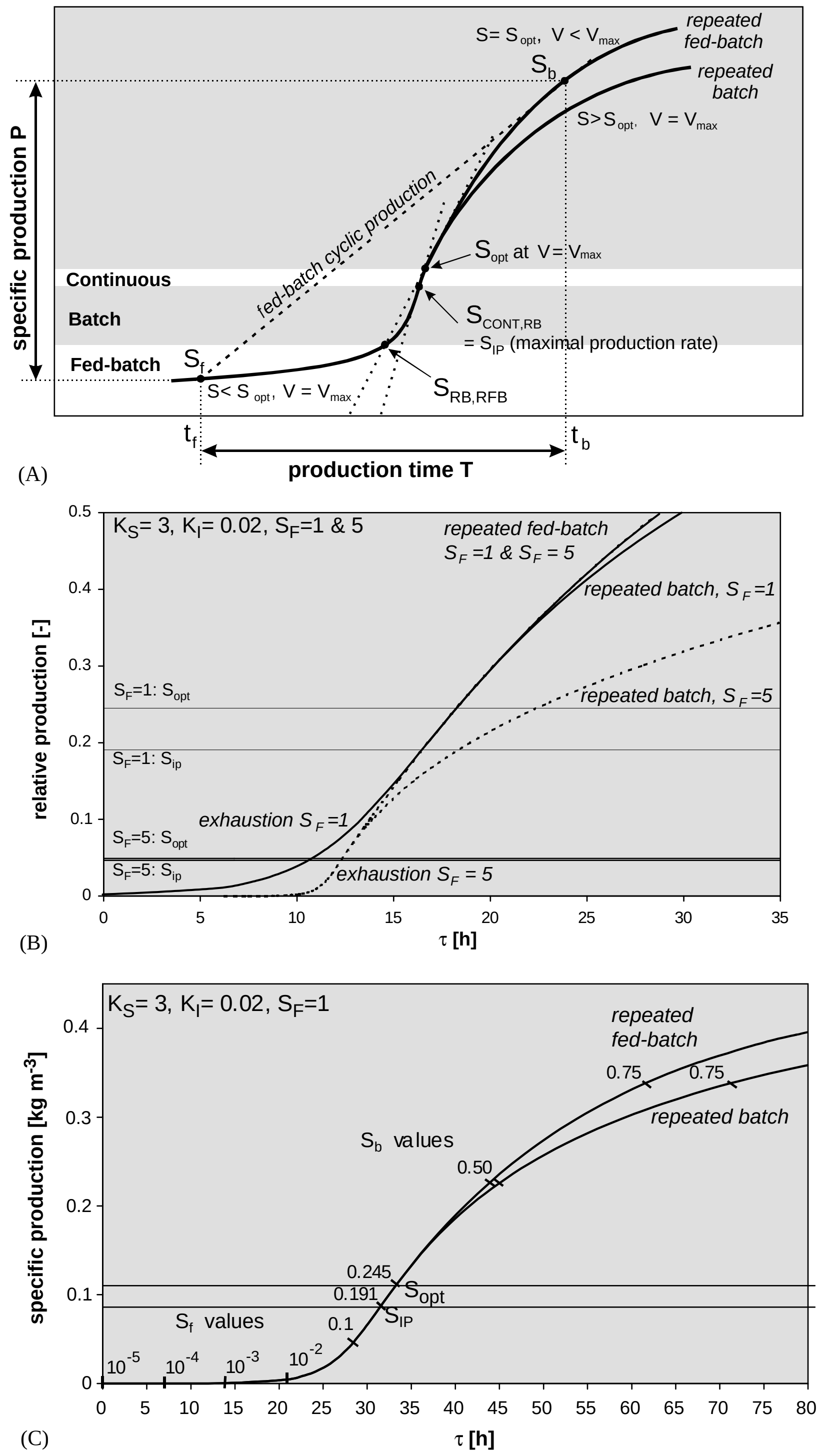

Fig. 6. Production Curves: (A) general curves for batch and fed-batch operation; (B) relative production for strong inhibited production ($K_S = 3$ (kg m^{-3}), $K_I = 0.02$ (kg m^{-3}), $\mu_{\max} = 1$ (h^{-1}), $Y = 0.45$) at $S_F = 1$ (kg m^{-3})) and $S_F = 5$ (kg m^{-3}); and (C) specific production curve for strong inhibited production at $S_F = 1$ (kg m^{-3}).

the initial and final substrate concentrations of the optimal cycle. S_f is the final state requirement, and S_b is realised by the proper residue ratio. In that case, the specific production rate equals $(P\{t_f\} - P\{t_b\})/(t_f - t_b)$. From the curve it follows that three areas of modes of operation can be distinguished. For this classification, final state values higher than S_{opt} fall outside the normal operating region. They are not realistic, because for $S_f > S_{\text{opt}}$, the production rate is less than S_{opt}, while hardly any exhaustion takes place.

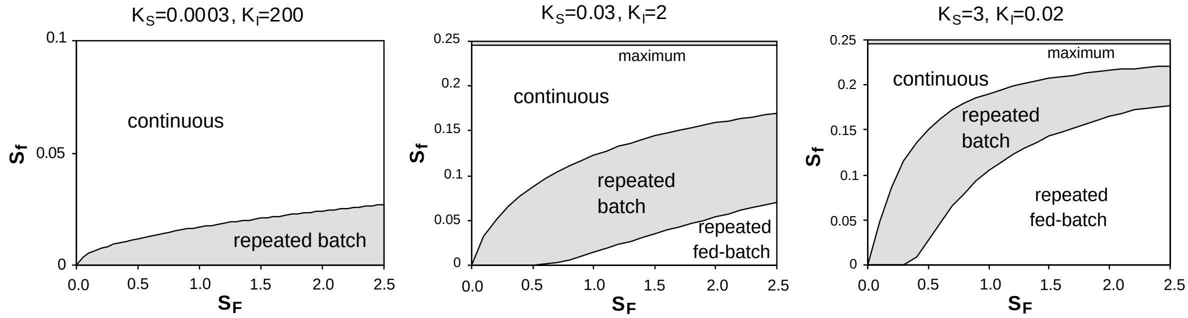

Fig. 7. The optimal mode of operation as a function of S_F and S_f for production of weak ($K_S = 0.3.10^{-3}$ (kg m^{-3}), $K_I = 200$ (kg m^{-3})), medium ($K_S = 0.03$ (kg m^{-3}), $K_I = 2$ (kg m^{-3})), strong inhibition ($K_S = 3$ (kg m^{-3}), $K_I = 0.02$ (kg m^{-3})).

• Continuous operation: $S_{\mathrm{CONT},RB} \leqslant S_f < S_{\mathrm{opt}}$: When S_f is larger than the inflection point S_{IP}, then continuous operation is optimal. Therefore, the transition point between continuous and batch operation, $S_{\mathrm{CONT},RB}$, corresponds to the inflection point S_{IP}, which can be found from the condition $\mathrm{d}^2P/\mathrm{d}t^2 = 0$, which is equal to $\mathrm{d}^2X/\mathrm{d}t^2 = 0$. From Eq. (1) it follows that this is satisfied if $\mathrm{d}(\mu\{S\}X)/\mathrm{d}t = 0$. Using Eq. (12), X can be eliminated and the following condition is obtained:

$$\frac{\partial X}{\partial S_f} = \frac{\partial}{\partial S_f}(\mu\{S_f\}Y(S_F - S_f)) = 0. \tag{19}$$

Then for the transition point, one can derive

$$S_f = S_{\mathrm{CONT},RB} = \frac{\sqrt{K_S(S_F + K_S + S_F^2/K_I)} - K_S}{1 + S_F/K_I}. \tag{20}$$

• Repeated batch operation: $S_{RB,RFB} \leqslant S_f < S_{\mathrm{CONT},RB}$: Repeated batch operation is optimal if S_f is located between the point where the tangent contacts the Production Curve at S_{opt} and the point S_{IP}. At S_{opt} the Production Curves of fed-batch and repeated fed-batch diverge. Thus, $S_{RB,RFB}$ can be determined from the condition that the tangent at S_{opt}, $\mathrm{d}P/\mathrm{d}t$, equals the production rate for the cycle S_{opt} to S_f, $(P\{S_f\} - P\{S_{\mathrm{opt}}\})/T$.

$$\left.\frac{P_{RB}}{T}\right|_{S=S_f} = \left.\frac{\mathrm{d}X}{\mathrm{d}t}\right|_{S=S_{\mathrm{opt}}}. \tag{21}$$

Consequently, for repeated batch operation S_b is between S_f and S_{opt}.

• Repeated fed-batch operation: $0 < S_f < S_{RB,RFB}$: In this case repeated fed-batch is the optimal mode of operation.

Fig. 7 shows the optimal modes of operation for three different levels of inhibition. The results are consistent with the division made in Table 1. A method to combine all control mode transitions for different kinetics is to plot the "degree of exhaustion" ε against the "degree of difficulty" of exhaustion δ, where ε and δ are defined as

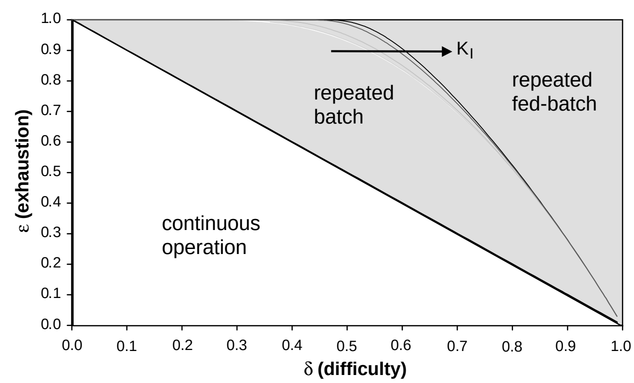

Fig. 8. Optimal mode of operations related to the degree of difficulty (δ) and the degree of exhaustion (ε) for five degrees of inhibition ((K_S (kg m^{-3}), K_I (kg m^{-3})) = (0.0003, 200), (0.003, 20), (0.03, 2), (0.3, 0.2), (3, 0.02).

$$\varepsilon = \frac{P}{P_{\max}}, \quad \delta = \frac{P_{\max} - P_{\max,\mathrm{characteristic}}}{P_{\max}}. \tag{22}$$

The degree of exhaustion ε is a quantitative measure indicating how much of the feed is converted or separated and the degree of difficulty is a qualitative measure describing how difficult it is to convert or to separate, these components. Usually, such a measure contains parameters for component properties, equipment capabilities, feed composition, and product requirements (Betlem & Roffel, 1997). This method has been applied earlier in a similar fashion for other cyclic processes such as batch distillation (Betlem, Krijnsen, & Huijnen, 1998). Both indicators are scaled between 0 and 1. $P_{\max,\mathrm{characteristic}}$ is the maximum production which can be realised under a certain control policy which is characteristic for the difficulty of the operation. In case the $P = P_{\max,\mathrm{characteristic}}$ then $\varepsilon + \delta = 1$. In case of biomass production ε and δ can be defined as:

$$\varepsilon = \frac{S_{\mathrm{opt}} - S_f}{S_{\mathrm{opt}}}, \quad \delta = \frac{S_{\mathrm{CONT},RB}}{S_{\mathrm{otp}}}. \tag{23}$$

S_{opt} at $V = V_{\max}$ in Fig. 6A can be interpreted as the transition point between the production phase during which

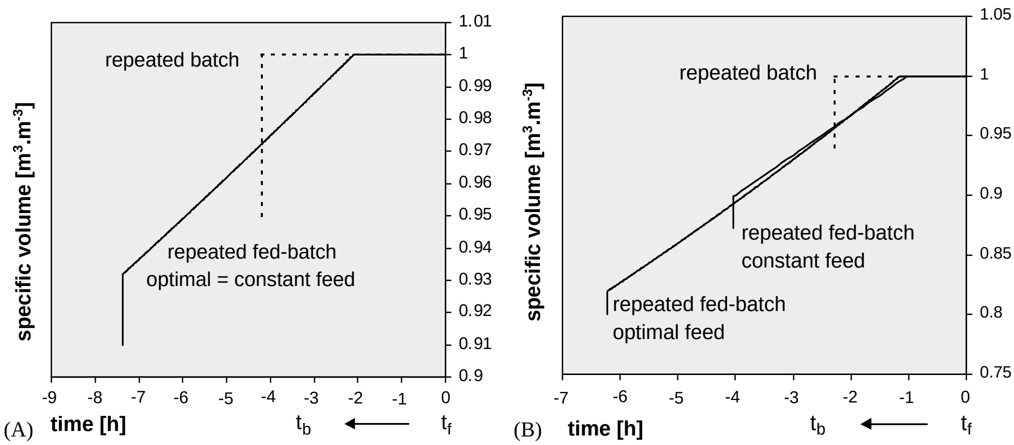

Fig. 9. Change of volume with time at the production rate maxima of repeated batch and optimal and suboptimal (constant feed rate) repeated fed-batch ($\mu_{\max} = 1$ (h^{-1}), $Y = 0.45$, $S_F = 1$ ($\mathrm{kg\ m^{-3}}$)) and $S_f = 0.01$ ($\mathrm{kg\ m^{-3}}$)). (A) weak inhibition (($K_S = 0.03$ ($\mathrm{kg\ m^{-3}}$), $K_I = 2$ ($\mathrm{kg\ m^{-3}}$)), and (B) strong inhibition (($K_S = 3$ ($\mathrm{kg\ m^{-3}}$), $K_I = 0.02$ ($\mathrm{kg\ m^{-3}}$)).

the biomass grows exponentially and the exhaustion phase during which the substrate is exhausted to the desired final condition. So, S_{opt} can be considered as the maximal realistic final substrate concentration, since a higher final concentration is always ineffective. The transition point between continuous and batch operation has been taken to characterise for the difficulty of the conversion. Fig. 7 shows that for a certain value of the substrate feed concentration (e.g. $S_F = 2\,[\mathrm{kg.m^{-3}}]$) the ratio between $S_{\mathrm{CONT},RB}$ and S_{opt} goes from a few percent for a relatively weak inhibition to nearly hundred percent for a strong inhibition. The parameter δ has been chosen such that $\varepsilon\{S_f = S_{\mathrm{CONT},RB}\} + \delta = 1$. The denominator of the right-hand term is a measure of the affinity of the biomass to the substrate concentration. The numerator reflects the influence of the inhibition and if $K_I \to \infty$, it becomes one.

Fig. 8 shows the optimal modes of operation as a function of exhaustion and conversion difficulty for five cases with the same S_{opt}. If K_I becomes larger and the exhaustion is high, the transition point between batch and fed-batch shifts to the right. For Monod kinetics, repeated fed-batch operation is not an option and Eq. (23) with $K_I \to \infty$, is sufficient to determine the optimal mode of operation.

6. Sub-optimal operation with recycles

Repeated fed-batch operation with optimal feed control can be approached by an operation with constant feed control, which is easier to implement. The procedure applied to the cyclic operation is the following. A finished batch is refreshed with S_F such that the growth rate at start-up equals $\mu_{\%}$ of $\mu_{\max}$. Next, the feed rate is set at:

$$F_{\mathrm{in}} = \mu_{\%}\mu\{S_{\mathrm{opt}}\}\eta V_{\max}. \tag{24}$$

During the conversion phase the substrate concentration will only decrease. First (see Fig. 2), the growth rate increases from $\mu_{\%}$ to $\mu_{\max}$. Next, it decreases and the feeding is stopped when the growth rate has returned to $\mu_{\%}$. The residue ratio, η, is used to ensure the final volume becomes $V_{\max}$. When $\mu_{\%}$ is taken as 100%, the reactor will be filled up such that at start-up: $V = V_{\max}$ and $S = S_{\mathrm{opt}}$. This agrees with the point where repeated fed-batch becomes repeated batch operation.

Fig. 4 shows the production rate for the suboptimal repeated fed-batch mode with constant feed rate. The production rate function has been calculated by simulations repeating the operation until a pseudo-stationary state becomes effective. The constant feed policy performs nearly as well as the optimal feed policy. For weak inhibition, it could be calculated that the maximum is reached at approximately $\mu_{\%} = 99.5\%$ and for strong inhibition the maximum was found at $\mu_{\%} = 98\%$. Fig. 9 shows the volume change in time for the three control modes at their maxima derived from Fig. 4. It appears that the maximum of the constant feed policy does not differ much from the maximum of the optimal policy. The maximum is approximately 2% less, however, it is more sensitive to the degrees of freedom. For weak inhibition, both are identical and for strong inhibition, the optimal cycle period of constant feed becomes shorter than for the optimal feed policy.

7. Discussion and conclusions

The biomass production differs in some respects to other cyclic processes. Typical characteristics of this process are:

- At cyclic operation, a direct relationship exists between the biomass and the substrate concentration.
- During the feeding phase of the optimal fed-batch control, the system is kept at maximum growth rate. The driving force of the system remains unchanged. Dynamic optimal control focuses on the future and tends to compensate a difficult exhaustion with a cautious start. In this case too, the dynamic optimal operation does not coincide with

momentary optimal operation, which occurs at a lower substrate concentration.
- As a result of the previous two points, the Production Curve is independent of the cycle time and only depends on the kinetic parameters.

A method to describe the optimal operation has been developed for bioreactions with substrate inhibition. Based on the Production Curve, the transition point from continuous to repeated batch and from repeated batch to repeated fed-batch with dynamic optimal feed rate have been determined. It has been shown that the optimal control mode can be described by the combination of a term that indicates the performance level (degree of difficulty) and a term representing the degree of exhaustion. Simulation studies show that fed-batch with constant feed is not much inferior to fed-batch with optimal feed control, because the residue ratio ensures that the substrate concentration will occur in the area of optimal growth. For strong inhibition the cycle time becomes shorter.

Notation

F_{in}	inflow rate reactor, $m^3\ h^{-1}$
F_{out}	outflow rate reactor, $m^3\ h^{-1}$
K_I	inhibition constant, $kg\ m^{-3}$
K_S	saturation (or Monod) constant, $kg\ m^{-3}$
P	specific production, $kg\ m^{-3}$
P_{batch}	P defined for batch operation, $kg\ m^{-3}$
$P_{\text{continuous}}$	P defined for continuous operation, $kg\ m^{-3}$
$P_{\max}$	maximal P, $kg\ m^{-3}$
$P_{\max,\text{characteristic}}$	maximal P for certain control strategy, $kg\ m^{-3}$
$P_{\text{potential}}$	maximal P according Eq. (8), $kg\ m^{-3}$
P_{relative}	relative production $P/P_{\text{potential}}$ [dimensionless]
PR	specific production rate, $kg\ m^{-3}\ h^{-1}$
S	substrate concentration, $kg\ m^{-3}$
T	cycle time, h
V	reactor volume, m^3
$V_{\max}$	maximum reactor volume, m^3
X	biomass concentration, $kg\ m^{-3}$
X_f	biomass product concentration, $kg\ m^{-3}$
Y	biomass yield [dimensionless]

Greek letters

δ	degree of difficulty of conversion (def.: Eq. (23))
ε	degree of exhaustion (def.: Eq. (23))
η	residue volume ratio [dimensionless]
μ	specific growth rate, h^{-1}
$\mu_{\max}$	maximum specific growth rate, h^{-1}
$\mu_{\%}$	percentage of $\mu_{\max}$, h^{-1}

Indices

0	start (after product removal)
CONT,RB	transition from continuous to repeated batch operation
b	begin batch (after refreshing)
f	final (after processing)
F	feed
IP	at inflection point
opt	at maximum growth rate
RB, RFB	transition from repeated batch to repeated fed-batch operation

References

Betlem, B. H. L., Krijnsen, H. C., & Huijnen, H. (1998). Optimal batch distillation control based on specific measures. *Chemical Engineering Journal*, *71*, 111–126.

Betlem, B. H. L., & Roffel, B. (1997). Integrated manufacturing of cyclic/batch processes. *ICSC-WMC 97*, Auckland, New Zealand (pp. 428–432).

Brown, W. A., & Cooper, D. G. (1991). Self-cycling fermentation applied to acinetobacter calcoceticus RAG-1. *Applied Environmental Microbiology*, *57*, 2901–2906.

Cazzador, L. (1988). On the optimal control of fed-batch reactors with substrate-inhibited kinetics. *Biotechnology and Bioengineering*, *31*, 670–674.

Chang, C. C., Ryu, D. D. Y., Park, C. S., & Kim, J. Y. (1998). Improvement of heterologous protein productivity using recombinant Yarrowia Lipolytica and cyclic fed-batch process strategy. *Biotechnology and Bioengineering*, *59*, 379–385.

Edwards, V. H. (1970). The influence of high substrate concentrations on microbial kinetics. *Biotechnology and Bioengineering*, *12*, 679–712.

Godin, F. B., Cooper, D. G., & Rey, A. D. (1999). Development and solution of a balance model applied to the SCF process. *Chemical Engineering Science*, *54*, 565–578.

Hasegawa, S., Matsubara, M., & Shimizu, K. (1987). Noninferior periodic operation of the metabolite producing biological reactor. *Biotechnology and Bioengineering*, *30*, 703–716.

Hughes, S. M., & Cooper, D. G. (1996). Biodegradation of phenol using the self-cycling fermentation (SCF) process. *Biotechnology and Bioengineering*, *51*, 112–119.

Keller, R., & Dunn, I. J. (1978). Computer simulation of the biomass production rate of cyclic fed batch continuous culture. *Journal of Applied Chemistry and Biotechnology*, *28*, 784–790.

Lee, T., Pham, M. Q., Weigand, W. A., Harvey, S. P., & Bentley, W. E. (1996). Bioreactor strategies for the treatment of growth-inhibitory waste. *Biotechnology Programming*, *12*, 533–539.

Matsubaru, M., Hasegawa, S., & Shimizu, K. (1985). Noninferior periodic operation of the biological reactor. *Biotechnology and Bioengineering*, *27*, 1214–1222.

Menawat, A., Muthurasan, R., & Coughanowr, D. R. (1987). Singular control strategy for a fed-batch bioreactor: Numerical approach *AIChE Journal*, *33*, 776–783.

Modak, J. M., & Lim, H. C. (1992). Optimal mode of operation of bioreactor for fermentation processes. *Chemical Engineering Science*, *47*, 3869–3884.

Modak, J. M., Lim, H. C., & Tayeb, Y. J. (1986). General characteristics of optimal feed rate profiles for various fed-batch fermentation processes. *Biotechnology and Bioengineering*, *28*, 1396–1407.

Palanki, S., Kravaris, C., & Wang, Y. H. (1993). Synthesis of state feedback laws for end-point optimization in batch processes. *Chemical Engineering Science*, *48*, 135–152.

Park, S., & Ramirez, W. F. (1988). Optimal production of secreted protein in fed-batch reactors. *AIChE Journal*, *34*, 1550–1558.

Parulekar, S. J. (1992). Analytical optimization of some single-cycle and repeated fed-batch fermentations. *Chemical Engineering Science*, *47*, 4077–4097.

Pirt, S. J. (1974). The theory of fetch-batch culture with reference in the penicillin fermentation. *Journal of Applied Chemistry and Biotechnology*, *24*, 415–424.

Rippin, D. W. T. (1983). Simulation of single- and multi-product batch chemical plants for optimal design and operation. *Computers and Chemical Engineering*, *30*, 137–156.

Roels, J. A. (1983). *Energetics and kinetics in biotechnology*. Amsterdam: Elsevier.

San, K. Y., & Stephanopoulos, G. (1984). A note on the optimality criteria for maximum biomass production in a fed-batch fermentor. *Biotechnology and Bioengineering*, *26*, 1261–1264.

San, K. Y., & Stephanopoulos, G. (1986). The effect of growth rate delays in substrate-inhibited kinetics on the optimal profile of fed-batch reactors. *Biotechnology and Bioengineering*, *28*, 356–361.

Shukla, P. K., & Pushpavanam, S. (1998). Optimisation of bio-chemical reactors. *Chemical Engineering Science*, *53*, 341–352.

Weigand, W. A. (1981). Maximum cell productivity by repeated fed-batch culture for constant yield case. *Biotechnology and Bioengineering*, *23*, 249–266.

Weigand, W. A., Lim, H. C., Creagan, C. C., & Mohler, R. D. (1979). Optimization of a repeated fed-batch reactor for maximum cell productivity. *Biotechnology Bioengineering Symposium*, *9*, 335–348.

Wincure, B. M., Cooper, D. G., & Rey, A. (1995). Mathematical model of self-cycling fermentation. *Biotechnology and Bioengineering*, *46*, 180–183.